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α-chymotrypsin in water–acetone and water–dimethyl sulfoxide mixtures: Effect of preferential solvation and hydration

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Abstract

© 2017 Wiley Periodicals, Inc. We investigated water/organic solvent sorption and residual enzyme activity to simultaneously monitor preferential solvation/hydration of protein macromolecules in the entire range of water content at 25°C. We applied this approach to estimate protein destabilization/stabilization due to the preferential interactions of bovine pancreatic α-chymotrypsin with water-acetone (moderate-strength H-bond acceptor) and water-DMSO (strong H-bond acceptor) mixtures. There are three concentration regimes for the dried α-chymotrypsin. α-Chymotrypsin is preferentially hydrated at high water content. The residual enzyme activity values are close to 100%. At intermediate water content, the dehydrated α-chymotrypsin has a higher affinity for acetone/DMSO than for water. Residual enzyme activity is minimal in this concentration range. The acetone/DMSO molecules are preferentially excluded from the protein surface at the lowest water content, resulting in preferential hydration. The residual catalytic activity in the water-poor acetone is ~80%, compared with that observed after incubation in pure water. This effect is very small for the water-poor DMSO. Two different schemes are operative for the hydrated enzyme. At high and intermediate water content, α-chymotrypsin exhibits preferential hydration. However, at intermediate water content, in contrast to the dried enzyme, the initially hydrated α-chymotrypsin possesses increased preferential hydration parameters. At low water content, no residual enzyme activity was observed. Preferential binding of DMSO/acetone to α-chymotrypsin was detected. Our data clearly demonstrate that the hydrogen bond accepting ability of organic solvents and the protein hydration level constitute key factors in determining the stability of protein–water–organic solvent systems.

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Keywords

bovine pancreatic α-chymotrypsin, dimethyl sulfoxide (DMSO), preferential solvation, protein hydration, protein stabilization

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